

This Page Is Inserted by IFW Operations
and is not a part of the Official Record

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

**As rescanning documents *will not* correct images,
please do not report the images to the
Image Problem Mailbox.**

THIS PAGE BLANK (USPTO)

I. K., 1992,
*f Biological*ith specific
f Biological
development
histological
xybutyratery blend of
Journal of
ospheres of
ulated levo-
y (Reading,
xybutyrate).

Effect of adding non-volatile oil as a core material for the floating microspheres prepared by emulsion solvent diffusion method

J.-H. LEE†, T. G. PARK‡, Y.-B. LEE§, S.-C. SHIN§ and
H.-K. CHOI†*

† College of Pharmacy, Chosun University Kwangju, 501-859, Korea

‡ Department of Biological Sciences, Korea Advanced Institute of Science and
Technology, Taejon, 305-701, Korea

§ College of Pharmacy, Chonnam National University, Kwangju, 500-757,
Korea

(Received 5 January 2000; accepted 24 April 2000)

Eudragit® microspheres, to float in the gastrointestinal tract, were prepared to prolong a gastrointestinal transit time. To enhance their buoyancy, non-volatile oil was added to the dispersed phase. When an oil component was not miscible with water, over 90% was entrapped within the microspheres and prolonged the floating time of the microspheres. Depending on the solvent ratio, the morphologies of the microspheres were different and the best result was obtained when the ratio of dichloromethane:ethanol:isopropanol was 5:6:4. As the isopropanol portion increased, the time to form microspheres was delayed and the amount of fibre-like substance produced was decreased, due to the slow diffusion rate of the solvent. Compared with microspheres prepared without non-volatile oil, the release rate of the drug from microspheres was faster in all cases tested, except the microspheres containing mineral oil. The solubility of the drug in the non-volatile oil affected the release profiles of the drugs. The non-volatile oil tends to decrease the glass transition temperature of prepared microspheres and change the release profile. The internal morphology of the microspheres was slightly different depending on the entrapped oil phase used. Tiny spherical objects were present at the inner surface of microspheres and the inside of the shell.

Keywords: Microspheres, floating, solvent ratio, release profile, morphology, non-volatile oil.

Introduction

To develop oral drug delivery systems, it is necessary to optimize both the residence time of the system in the gastrointestinal tract and the release rate of the active ingredient from the system. Only a limited number of approaches have been pursued to extend the gastric residence time (GRT) of a system. The efforts to extend GRT of an oral drug delivery system include the use of a passage delaying agent, mucoadhesive systems (Leußen *et al.* 1994), and floating devices (Shozo *et al.* 1988, Kawashima *et al.* 1991). Bechaard and Ladefoged (1978) reported that

* To whom correspondence should be addressed: e-mail: hgchoi@chosun.ac.kr

Volume 18 Number 1 January–February 2001

ISSN 0265-2048

JOURNAL OF **MICROENCAPSULATION**

MICROCAPSULES LIPOSOMES NANOPARTICLES MICROCELLS MICROSPHERES

The official journal of the International Microencapsulation Society

Univ. of Minn.
Bio-Medical
Library

01 23 01



the density was more important than the size to modify the transit time of multiple units. Mori *et al.* (1989) and Christensen *et al.* (1985) reported that the small intestinal transit time of granules was not influenced by specific gravity; however, the gastric emptying rate was changed, depending on the specific gravity of the granules. Kawashima *et al.* (1991) proposed drug loaded hollow microspheres (microballoons) to prolong GRT of the dosage form. Atyabi *et al.* (1994) evaluated ion exchange resins that float by producing CO_2 . Recently, Iannuccelli *et al.* (1998a, 1998b) reported that an air compartment multiple-unit system showed excellent buoyancy *in vitro* and prolonged GRT over the control *in vivo* in the fed state after a meal and after a succession of meals. However, in the fasted state, the intragastric buoyancy of the devices did not influence its GRT.

In a previous study (Lee *et al.* 1999), an improved method was reported to prepare floating microspheres using acrylic resin. In the present study, the effects are investigated of adding a non-volatile oil as a core material on the characteristics of acrylic microspheres, such as floating time, morphology, drug loading efficiency, and release profile of the drugs. It was expected that non-volatile oil may improve the floating behaviour of the microspheres. Also, it may enhance the absorption rate of cyclosporin A when appropriate vegetable oil is entrapped together. The results were compared with the conventional o/w solvent diffusion and evaporation method used in a previous study. Three kinds of low-density, non-volatile oils, mineral oil (MO), isopropyl myristate (IPM), and Labrafil® 1944, were tested.

Materials and methods

Materials

Eudragit® S100 was a gift from Röhm Pharma Co. (Darmstadt, Germany). Ketoprofen, tenoxicam, piroxicam, and tacrine HCl were obtained from Jeil Pharmaceutical Co. (Seoul, Korea). Cyclosporin A was obtained from Jeil Jedang Research Labs. (Seoul, Korea). Mineral oil, isopropyl myristate, and poly (vinyl alcohol) were purchased from Sigma Chemicals (St. Louis, MO, USA) and Labrafil® 1944 was purchased from Gattefossé Korea (Seoul, Korea). All other chemicals were reagent grade and were used as received without further purification.

Preparation of microspheres

Microspheres containing non-volatile oil as a core material were prepared by a solvent evaporation method. Ethanol, isopropyl alcohol, dichloromethane, and non-volatile oil were mixed with various mixing ratios. Two hundred milligrams of a selected drug and 1 g of Eudragit® S100 were dissolved in 10 ml of the solvent mixture. The polymer solution was slowly introduced into 1000 ml of 0.4% (w/v) poly (vinyl alcohol) aqueous solution while being stirred at 250 rpm using a mechanical stirrer (RZR 2000, Cafrimo Co., Canada) equipped with a three-bladed propeller at room temperature. The solution was stirred for 10 min and the microspheres were collected by filtration. The collected microspheres were dried for 12 h at 50°C.

Particle size

Prepared with stirring and particle scattering (1)

Yield of microspheres

The pre-weighed. T volatile con-

Determination of particle size

Microparticle size. The diluted detector (SI) was calculated to that of the volatile oil by measuring 1

Thermal analysis

To measure scanning ca flow of 35 n

Drug release

The drug release Inst., Korea of the dissolution maintained aliquot was the drug re

Floating time

An in a dispersing dispersing : 100 rpm. E down was weighed aft

Morphology

The microspheres (JMS 840-distribution

of multiple the small ; however, vity of the crospheres) evaluated celli *et al.* m showed in the fed d state, the

eported to the effects racteristics ; efficiency, ay improve absorption ether. The evaporation olatile oils, e tested.

Germany). 1 from Jeil 1 Jeiljedang poly (vinyl USA) and). All other out further

Particle size analysis

Prepared microspheres were dispersed in 0.02% aqueous tween 80 solution with stirring and ultrasonification to disperse the microspheres. Mean particle size and particle size distribution of the microspheres were determined by laser light scattering (SALD 2001, Shimadzu, MD).

Yield of microsphere formation

The prepared microspheres with a size range of 75–1000 μ m were collected and weighed. The measured weight was divided by the total amount of all the non-volatile components used for the preparation of the microspheres.

Determination of loading efficiency

Microspheres prepared with 200 mg of a model drug were dissolved in ethanol. The diluted sample solution was analysed by the HPLC system with an UV detector (Shimadzu Scientific Instruments Inc., MD, USA). Loading efficiency was calculated by comparing the amount of the drug used to prepare microspheres to that of the drug loading into microspheres. The loading efficiency for the non-volatile oil was determined by dissolving them in pH 7.4 phosphate buffer and measuring the volume of the oil in the upper phase.

Thermal analysis of microspheres

To measure the glass transition temperature of microspheres, a differential scanning calorimeter (Mettler TA3000 system; open pan system in nitrogen gas flow of 35 ml/min; heating rate 10 K/min) was used.

Drug release test

The drug release test was carried out using a dissolution tester (DST, Fine Sci. Inst., Korea). Microspheres containing the active ingredient were placed in 500 ml of the dissolution medium at pHs of 2.0, 6.8, and 7.4. The temperature was maintained at 37°C. The medium was stirred at 100 rpm using a paddle, and an aliquot was withdrawn at pre-determined time intervals to analyse the amount of the drug released from microspheres by HPLC.

Floating test

An *in vitro* floating test was carried out using simulated gastric fluid as a dispersing medium. Microspheres were spread over the surface at 500 ml of dispersing medium at 37°C. The medium was agitated by a paddle rotating at 100 rpm. Each fraction of microspheres floating on the surface and those settled down was collected at a pre-determined time point. The collected sample was weighed after drying.

Morphology

The morphology was examined by light and scanning electron microscopy (JMS 840-A, JEOL, Japan). To investigate the internal morphology and the distribution of non-volatile oil, microspheres were divided into two pieces. One

piece was immersed in dichloromethane to solubilize the non-volatile oil and another piece remained intact. Well-dried half and whole microspheres were put on the sub and coated, under vacuum, with gold.

Results and discussion

Effect of solvent ratio and oil incorporation

To find an optimum solvent composition for the dispersed phase, the effect of various solvent ratios on the morphology of microspheres was investigated. Table 1 shows the effect of solvent ratio on the mean particle size of the microspheres. When the amount of dichloromethane or isopropyl alcohol was increased, the size of the microspheres was increased as expected (Lee *et al.* 1999). When ethanol, isopropanol, and dichloromethane were used as mixed solvents for the dispersed phase to dissolve the acrylic polymer in o/w solvent diffusion method, ethanol could preferentially diffuse out into aqueous medium, followed by isopropanol, due to their differential miscible with water. On the other hand, dichloromethane is sparsely miscible with water and diffused out more slowly and evaporated after the microspheres were nearly solidified (Kawashima *et al.* 1989). It was reported that a slower diffusion rate of the volatile solvents resulted in larger microspheres (Lee *et al.* 1999). Based on the results from the o/w solvent diffusion method, the solvent composition of dichloromethane:ethanol:isopropanol = 5:8:2 was used. The resultant microspheres were distorted, and many fibre-like structures were formed. It seemed that the addition of non-volatile oil shortened the time for the polymer to solidify, hence, there was less time to form a stable emulsion. To provide more time to form stable emulsion droplets, a composition with a higher content of isopropanol was used. The solvent ratio of 5:6:4 seemed to be appropriate for this study and was used in all other subsequent experiments. When the content of isopropanol was higher than the optimum composition, it showed a broader size distribution, non-spherical shape, and some of them collapsed.

Figure 1 shows the effect of including non-volatile oil as a core material on the size distribution of the microspheres. The mean particle size of the prepared microspheres containing non-volatile oil was similar to or larger than that without oil. This may be due to a slightly larger emulsion droplet size generated after the evaporation of volatile components when compared to the case without non-

Figure 1. E
compo:
isoprop
contain

volatile oil.
than that w
generated a
without nor
a broader si

The yiel
non-volatile
like aggrega
higher than
shapes that

Effect of no

Three k
used in this
dispersed p
shiny and t
containing
WO-MS (f
together an
and dense,;
as can be se
outer surfa
quick solidi

Table 1. Effect of various non-volatile oils and solvent ratios on the mean particle size and the yield of microsphere formation.

Solvent ratio (D:E:I)	Mineral oil		Labrafil 1944		Isopropyl myristate	
	Size (μm) (Mean ± SD)	Yield (%)	Size (μm) (Mean ± SD)	Yield (%)	Size (μm) (Mean ± SD)	Yield (%)
5:8:2	277.5 ± 17.2	91.9	296.1 ± 18.3	89.9	254.7 ± 4.42	84.0
5:6:4	349.9 ± 9.09	93.0	348.3 ± 9.82	89.2	354.2 ± 8.58	89.2
5:4:6	388.9 ± 37.2	92.0	394.7 ± 27.5	93.9	391.7 ± 17.5	93.2
5:2:8	413.2 ± 43.8	93.6	412.6 ± 20.3	92.2	397.0 ± 22.5	94.3

D, dichloromethane; E, ethanol; I, isopropyl alcohol.

ile oil and
es were put

the effect of
ed. Table 1
icrospheres.
ed, the size
en ethanol,
e dispersed
od, ethanol
sopropanol,
romethane
orated after
as reported
icrospheres
method, the

was used.
ctures were
time for the
ulsion. To
ith a higher
med to be
periments.
position; it
e of them

aterial on the
e prepared
at without
ed after the
ithout non-

icle size and

	Yield (%)
42	84.0
58	89.2
7.5	93.2
2.5	94.3

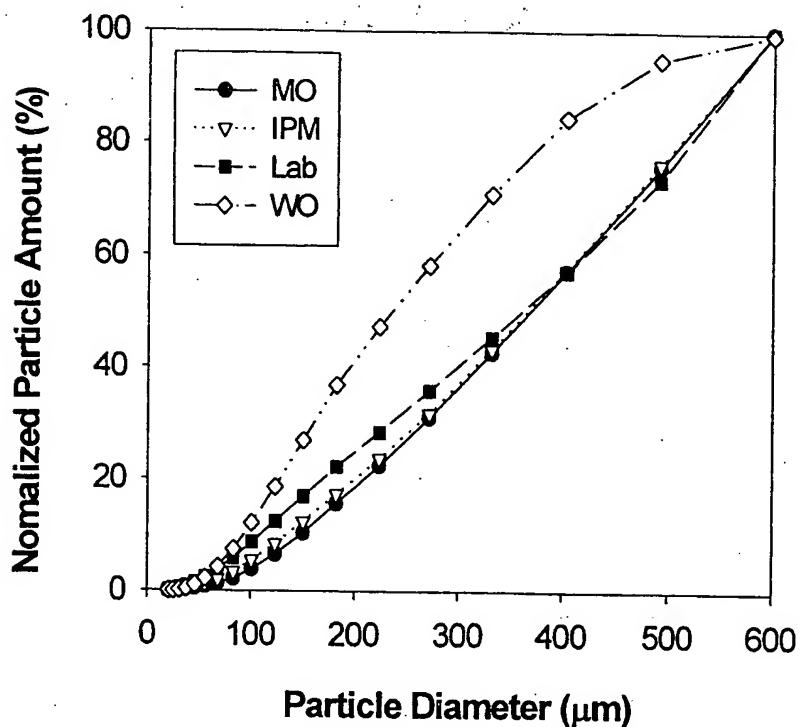


Figure 1. Effect of non-volatile oil on the size distribution of microspheres. The solvent composition used to prepare the microspheres was dichloromethane:ethanol:isopropanol = 5:6:4 (MO, microspheres containing mineral oil; IPM, microspheres containing isopropyl alcohol, Lab, microspheres containing Labrafil® 1944).

volatile oil. The microspheres containing non-volatile oil was similar to or larger than that without oil. This may be due to a slightly larger emulsion droplet size generated after the evaporation of volatile components when compared to the case without non-volatile oil. The microspheres containing non-volatile oil also showed a broader size distribution.

The yield of microsphere formation was higher than 80%, and over 90% of the non-volatile oil was entrapped. The addition of oil seemed to produce more fibre-like aggregates and it was aggravated when the amount of non-volatile oil was higher than 50%. The fibre-like aggregates produced were broken into small rod shapes that were difficult to separate from the prepared microspheres.

Effect of non-volatile oils on morphology

Three kinds of non-volatile oils, mineral oil, IPM, and Labrafil® 1944, were used in this study. The microspheres prepared with mineral oil (MO-MS) in the dispersed phase looked transparent. The surface of the microspheres was oily and shiny and they tended to stick together. The surface morphology of microspheres containing IPM (IPM-MS) or Labrafil® 1944 (Lab-MS) was similar to that of WO-MS (figure 2(a)). The surface looked opaque and white. They did not stick together and flowed freely. The outer surface of the microsphere shell was smooth and dense, and the internal part of the microsphere wall had more and larger pores, as can be seen in figure 2(b) and (c). Crotts and Park (1995) also reported that the outer surface, which contacts with water first, was smooth and dense due to its quick solidification. Mineral oil was not compatible with acrylic polymer and the

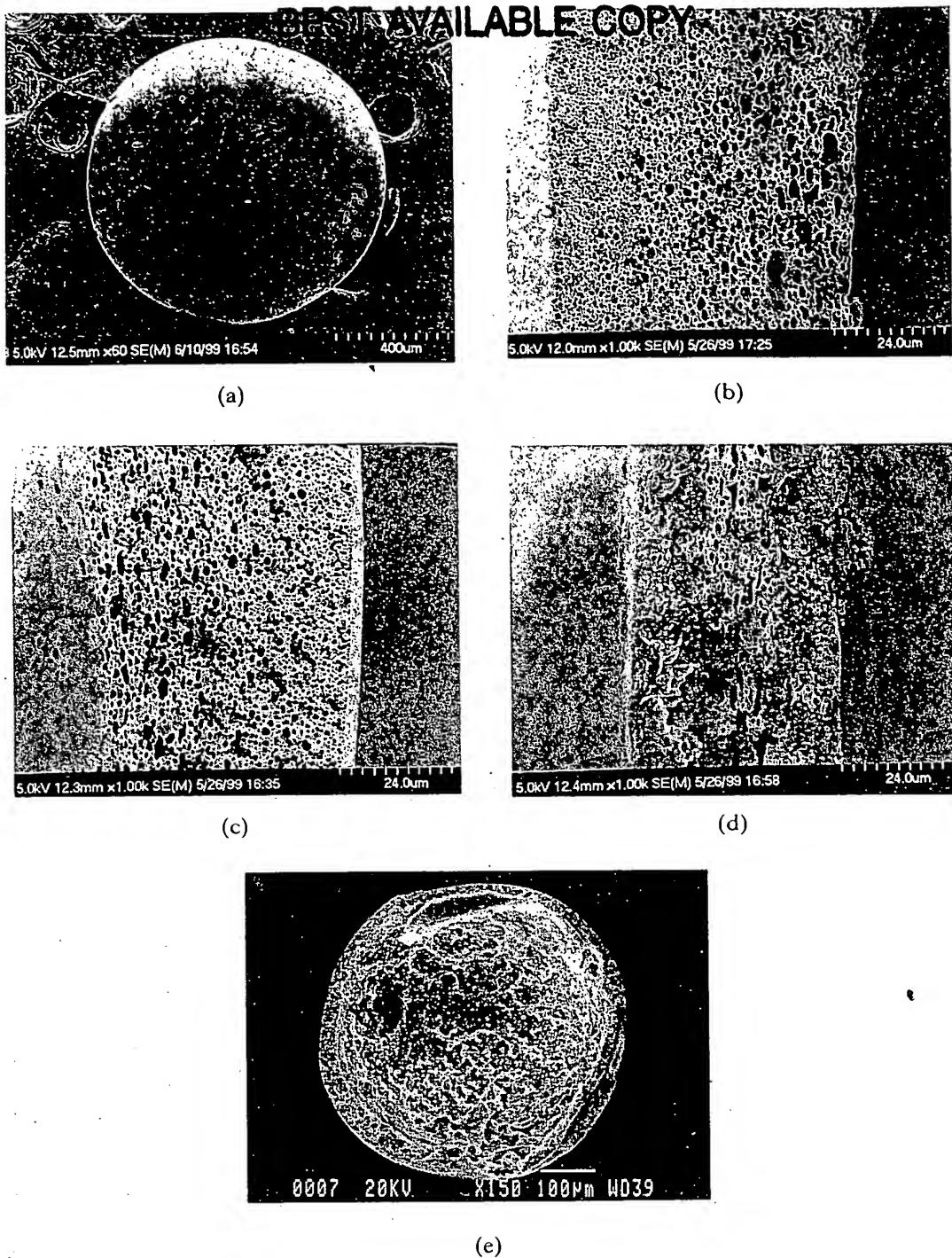


Figure 2. Scanning electron micrographs; (a) microsphere prepared with isopropyl myristate, (b), (c), (d) wall structure of microspheres containing isopropyl myristate, Labrafil® 1944, and mineral oil, respectively, and (e) internal view of microsphere showing many tiny spherical objects.

two phases were separated when the mixed solution in organic solvents was cast into a film. As a result, mineral oil was found inside and outside of the shell and filled the pores and channels within the shell of the microspheres. On the other hand, IPM or Labrafil® 1944 seemed to be compatible with acrylic polymer, and were not separated from the polymer when cast into a film. The microspheres

Figure 3.
The s
ethane

containing
2(a), (c) an
Micros
structure
appeared
(figure 2(e
was divide
other was
concluded
could not l

Floating te

The ad
The buoy
results fro
within the
from invad
floating on
showed tha
Labrafil® 1
did not in
entrapped

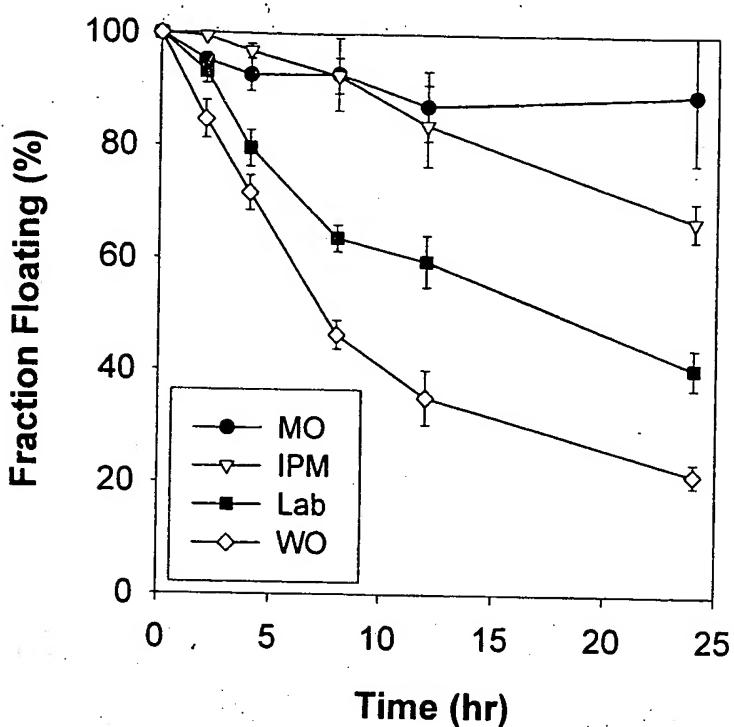


Figure 3. Floating behaviour of microspheres without a drug in a simulated gastric fluid. The solvent composition used to prepare the microspheres was dichloromethane: ethanol:isopropanol = 5:6:4.

containing mineral oil had a thinner and less porous wall than the others (figures 2(a), (c) and (d)).

Microspheres containing non-volatile oil showed a characteristic internal structure when compared with that of WO-MS. Many tiny spherical objects appeared at the internal surface of the microspheres and/or inside the wall (figure 2(e)). To determine the nature of the spherical objects, the microsphere was divided into two pieces. One piece was washed with dichloromethane and the other was kept intact, and the samples were compared using SEM. It was concluded that they were not oil droplets but acrylic polymer, because they could not be removed by treating with dichloromethane.

Floating test

The addition of non-volatile oil improved the floatability of the microspheres. The buoyancy of microspheres containing non-volatile oil in aqueous medium results from a low density, due to not only a hollow structure but oil entrapped within the core and the shell of microspheres. In addition, the oil protected water from invading into the microspheres. Figure 3 shows the fraction of microspheres floating on the water with time. Microspheres containing mineral oil (MO-MS) showed that the best floatability followed IPM-MS, Lab-MS, and WO-MS. Since Labrafil® 1944, a surfactant, improves the wettability of the microsphere surface, it did not improve the floatability as much as IPM or mineral oil. No drug was entrapped into the microspheres used in this section of the study.

with isopropyl
propyl myristate,
f microsphere

ents was cast
the shell and
On the other
polymer, and
microspheres

Table 2. Solubility and loading efficiency (LE) of drugs within the microspheres containing non-volatile oil.

Drug	Mineral oil		IPM		Labrafil 1994		WO
	Solubility (mg/ml)	LE (%) (Mean \pm SD)	Solubility (mg/ml)	LE (%) (Mean \pm SD)	Solubility (mg/ml)	LE (%) (Mean \pm SD)	LE (%) (Mean \pm SD)
Cyclosporin	1.08	97.3 \pm 0.4	14.46	99.0 \pm 0.2	95.50	96.6 \pm 0.2	98.0 \pm 0.2
Ketoprofen	0.19	79.6 \pm 0.7	15.40	87.3 \pm 0.4	53.20	87.2 \pm 0.6	81.0 \pm 2.0
Piroxicam	0.02	73.7 \pm 0.9	0.32	69.8 \pm 0.4	2.59	75.3 \pm 0.4	75.7 \pm 0.2
Tenoxicam	0.01	60.4 \pm 1.6	0.22	57.0 \pm 1.0	0.78	51.6 \pm 0.4	63.5 \pm 2.1
Tacrine HCl	0.03	15.8 \pm 0.8	0.15	15.0 \pm 1.0	0.25	21.0 \pm 1.0	21.7 \pm 0.4

IPM, Isopropyl myristate, WO, Without oil

Effect of the solubility of drug in non-volatile oil on the loading efficiency

Generally, the loading efficiency is influenced by the relative solubility of a drug between a dispersed phase and a continuous phase. As shown in table 2, the addition of non-volatile oil did not improve the loading efficiency unless the drug had a high solubility in the selected oil. In the case of ketoprofen, microspheres containing IPM or Labrafil® 1994 showed improved loading efficiency, due to its higher solubility in those oil components. Piroxicam and tenoxicam have low solubilities in all additives, and their loading efficiencies were lower than that of WO-MS. Therefore, if the drug is soluble in an appropriate non-volatile oil, this method can be used to improve the loading efficiency.

Effect of pH and non-volatile oil on the release profile of ketoprofen

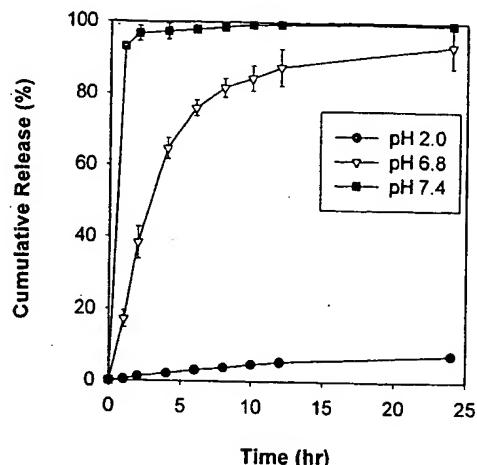
The effect of pH and non-volatile oil on the release profile of a drug was investigated using ketoprofen as a model drug. The release profiles of ketoprofen from the acrylic microspheres containing oil at pH 2.0, 6.8 and 7.4 are shown in figure 4. More than 90% of the drug was released within 1 h at pH 7.4, and less than 10% was released at pH 2.0, due to the enteric nature of the acrylic polymer. At pH 7.4, the drug release rates were similar, irrespective of the kind of non-volatile oil used, since the drug release rate was mainly controlled by polymer dissolution. The shell of the Eudragit microsphere was quickly dissolved in alkaline solution before the oil or drug could diffuse out of the microspheres. At pH 6.8, the microspheres containing oil tend to release the drug more quickly than WO-MS, except for MO-MS. When the polymer shell contacts aqueous medium, entrapped oil can escape easily and the drug solubilized in the oil can diffuse out into the release medium together. Labrafil® 1994 showed the highest solubility of ketoprofen among tested oils and showed the fastest release rate followed by IPM. The release profile of Lab-MNS at pH 6.8 was similar to that of pH 7.4. In addition, oil containing microspheres have a less dense and more porous shell than WO-MS, thus the aqueous release medium penetrates pores and channels created by oil that diffused out from the pores.

Non-volatile oil in the polymer shell of microspheres may play a role as a plasticizer. Figure 5 shows that the glass transition temperature (T_g) of microspheres containing non-volatile oil is lower than that of WO-MS, except for MO-

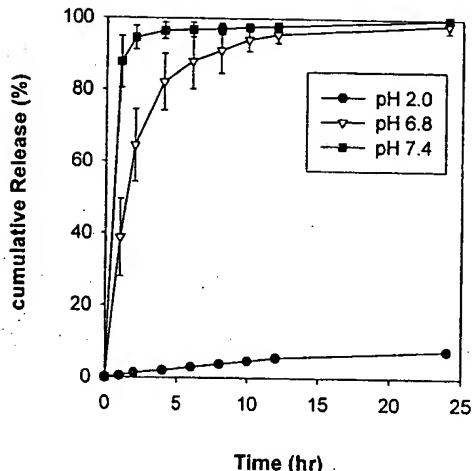
Figure 4.
volati
The
ethan

ospheres

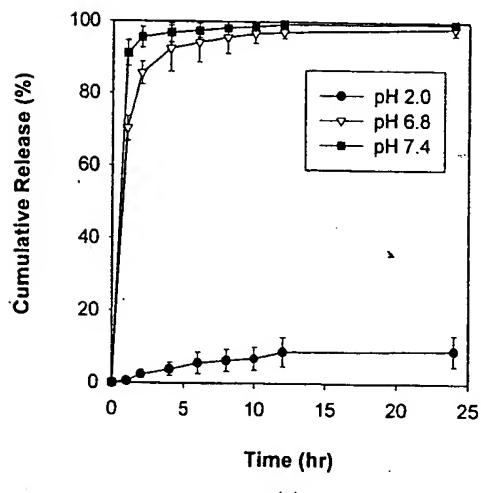
WO

LE (%)
(Mean \pm SD)98.0 \pm 0.2
81.0 \pm 2.0
75.7 \pm 0.2
63.5 \pm 2.1
21.7 \pm 0.4

(a)



(b)



(c)

Figure 4. The release profiles of ketoprofen from acrylic microspheres containing non-volatile oil at pH 2.0, 6.8, and 7.4. (a) Mineral oil, (b) IPM, and (c) Labrafil[®] 1944. The solvent composition used to prepare the microspheres was dichloromethane: ethanol:isopropanol = 5 : 6 : 4.

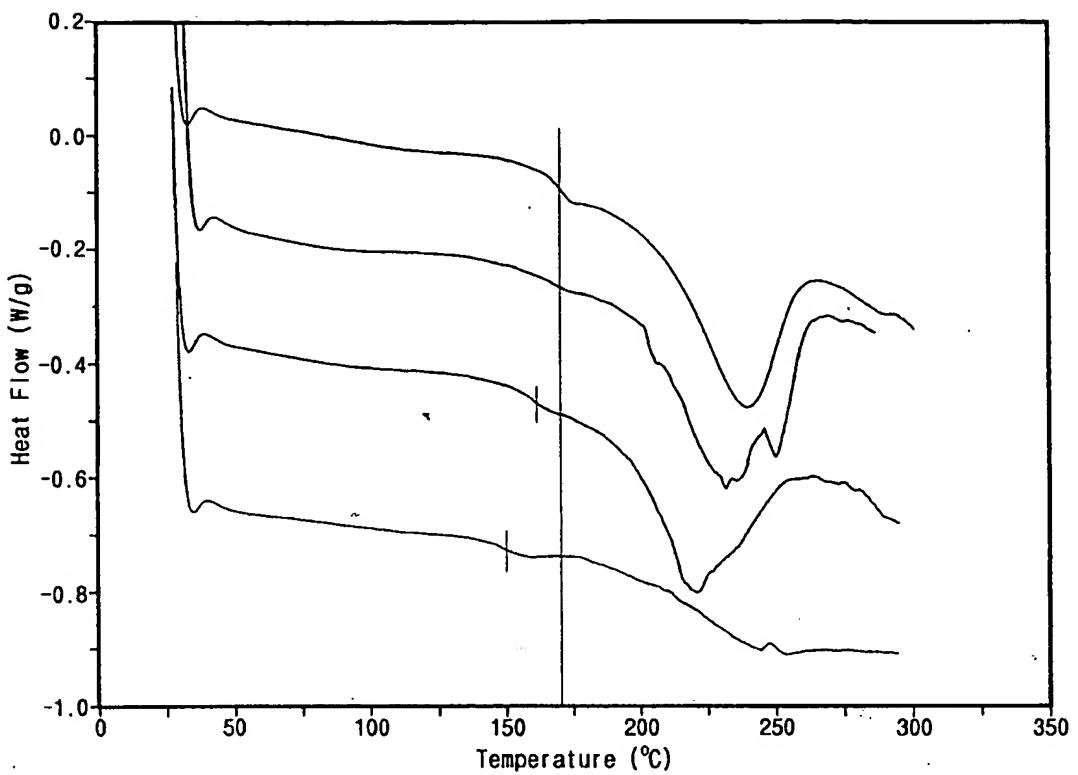


Figure 5. Differential scanning calorimetry thermograms of acrylic microspheres. From the top; microspheres without non-volatile oil (T_g : 169.4°C), microspheres containing mineral oil (T_g : 166.9°C), microspheres containing isopropyl myristate (T_g : 161.3°C), and microspheres containing Labrafil® 1944 (T_g : 148.4°C).

CHRISTENSEN
WIL
tran:
91-9
CROTT, G.
poly
IANNUCCELL
mul:
Inte:
IANNUCCELL
mul:
Inte:
KAWASHIMA
Pre:
by:
Scie:
KAWASHIMA
mul:
tran:
vivo:
LEE, J. H.,
usin:
LUEBEN, H.
and:
pep:
MORI, M.,
IMA:
the:
SHOZO, M.,
rele:
Che:

MS, indicating that IPM and Labrafil® 1944 are acting as plasticizers for the acrylic polymer. The release rate of MO-MS is similar to or slightly slower than that of WO-MS because mineral oil is a poor solvent for ketoprofen and cannot play a role as a plasticizer due to its incompatibility with the polymer.

Acknowledgements

This work was supported by the research grant from the Korea Science and Engineering Foundation (KOSEF 971-0713-106-2).

References

ATYABI, F., SHARMA, H. L., MOHAMMAD, H. A. M., and FELL, J. T., 1994, A novel floating system using ion exchange resins. *Proceedings of the International Symposium on Controlled Release of Bioactive Materials*, 21, 806-807.

BECHAARD, H., and LADEFOGED, K., 1978, Distribution of pellets in the gastrointestinal tract. The influence on transit time exerted by the density or diameter of pellets. *Journal of Pharmacy and Pharmacology*, 30, 690-692.

CHRISTENSEN, F. N., DAVIS, S. S., HARDY, J. G., TAYLOR, M. J., WHALLEY, D. R., and WILLSON, C. G., 1985, The use of gamma scintigraphy to follow the gastrointestinal transit of pharmaceutical formulation. *Journal of Pharmacy and Pharmacology*, **37**, 91-95.

CROTT, G., and PARK, T. G., 1995, Preparation of porous and nonporous biodegradable polymeric hollow microspheres. *Journal of Controlled Release*, **35**, 95-105.

IANNUCCELLI, V., COPPI, G., SANSONE, R., and FEROLLA, G., 1998a, Air compartment multiple-unit system for prolonged gastric residence. Part I. Formulation study. *International Journal of Pharmaceutics*, **174**, 47-54.

IANNUCCELLI, V., COPPI, G., SANSONE, R., and FEROLLA, G., 1998b, Air compartment multiple-unit system for prolonged gastric residence. Part II. *In vivo* evaluation. *International Journal of Pharmaceutics*, **174**, 55-62.

KAWASHIMA, Y., NIWA, T., HANADA, T., TAKEUCHI, H., IWANOTO, T., and ITOH, K., 1989, Preparation of controlled-release microspheres of ibuprofen with acrylic polymers by a novel quasi-emulsion solvent diffusion method. *Journal of Pharmaceutical Science*, **78**, 68-72.

KAWASHIMA, Y., NIWA, T., TAKEUCHI, H., HINO, T., and ITO, Y., 1991, Preparation of multiple unit hollow microspheres (microballoons) with acrylic resin containing tranilast and their drug release characteristics (*in vitro*) and floating behavior (*in vivo*). *Journal of Controlled Release*, **16**, 279-290.

LEE, J. H., PARK, T. G., and CHOI, H. K., 1999, Development of oral drug delivery system using floating microspheres. *Journal of Microencapsulation*, **16**, 715-729.

LUEBEN, H. L., LEHR, C.-M., RENTEL, C.-O., NOACH, A. B. J., DEBOER, A. G., VERHOEF, J. C., and JUNGINGER, H. E., 1994, Bioadhesive polymers for the peroral delivery of peptide drugs. *Journal of Controlled Release*, **29**, 329-338.

MORI, M., SHIRAI, Y., UEZONO, Y., NAKAMURA, Y., MAKITA, H., NAKANISHI, Y., and IMASATO, Y., 1989, Influence of specific gravity and food on movement of granules in the gastrointestinal tract of rats. *Chemical and Pharmaceutical Bulletin*, **37**, 738-741.

SHOZO, M., YAMAGUCHI, H., YOKOUCHI, C., TAKATA, M., and HON, W.-M., 1988, Sustained-release and intragastric-floating granules of indomethacin using chitosan in rabbits. *Chemical and Pharmaceutical Bulletin*, **36**, 4033-4038.